Abstract

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Title of Doctoral Thesis: Study of biological properties of hyaluronic acid derivatives

This PhD thesis, is dealing with physiological properties of Hyaluronic Acid labelled with ^{111}In isotope. HA is naturally occurring Glycosaminoglycan within organisms with many important functions on the cellular and organ level, including physiological and pathological processes. Its character of physiologically occurring substance makes its tracing in the organism very difficult. Thus preparation of suitable derivative and repeatable procedure of labelling of this molecule became the most important point of the whole work.

During the experimental part DTPA-HA molecule created by CPN Dolni Dobrouč has been labelled using well-known method used for labelling of DTPA molecules with ^{111}In. A reproducible procedure has been created where 0,4 M Acetate buffer pH 5,5 is mixed with an equal volume of DTPA-HA purified by gel filtration and required amount of isotope ^{111}In and the whole mixture is mixed for 0,5 h. This method was used for labelling of DTPA-HA molecules of three different molecular weights 10, 100 and 400kDa, which proved to be sufficiently stable and having properties of native HA.

Newly created HA 10, 100 and 400 were used for biodistributional, pharmacokinetic and elimination studies and for liver perfusion in situ.

Biodistribution revealed the same behaviour of all three examined molecular weights. A rapid uptake by liver from circulation and slow elimination from organism appeared. With higher molecular weight uptake was more quantitative.

Two-compartment model was used for calculation of pharmacokinetic parameters. Results show fast biodistribution phase and slow elimination phase, where with increasing molecular weight the volume of central compartment and elimination half-life grows and on the contrary distribution half-life and plasma clearance are getting lower.

Elimination of all three molecules was mostly renal with only small contribution of GIT. HPLC analysis of Mw of HA recovered from urine corresponded to low molecular weight fragments. Only for HA 10 a peak appeared with the same elution time as for the standard.

A huge amount of activity was taken up by liver within first ten minutes of liver perfusion. The following phase corresponded to the first order kinetics.

The results obtained were used for further research in the Radio-immunological Laboratory of Faculty of Pharmacy, Charles University, and for further research in collaboration with the Contipro C a. s.,, Dolni Dobrouč, which supported the conduct of this research.